REMARKS

Claim Amendments

Claims 114, 122, 130, 138, 146, 148, 150, 190, 199, 208, 217, 226, 229 and 232 have been amended to specify that the reference RNA sequence and the (selected) target viral RNA sequence are of similar length. These claims have also been amended to specify that the reference RNA sequence and the (selected) target viral RNA sequence can be amplified and detected by the same oligonucleotides. Support for the former amendment can be found in the disclosure in the present application in which the maxigene construct differs from the target viral RNA sequence by about 20 bases (see page 6, lines 19-21). Support for the latter amendment can be found at page 6, lines 15-18.

Claims 152-189 have been canceled.

New claims 235-248 have been added to specify that the multibase insert or the sequence not present in the selected target viral RNA sequence is about 21 nucleotides in length. Support for this language can be found in the length of an example of a multibase insert for HIV set forth at page 6, lines 24-29.

Applicants submit that these amendments do not constitute new matter and their entry is requested.

Summary of the Claims

The claims comprise two sets of claims, each directed to one of the reference RNA sequences. These sets of claims are summarized as follows.

Claims 114-151 and 235-241 are directed to processes for quantitation of a target viral RNA sequence in a sample, an amplification reaction mixture (claims 146-147 and 239), a reverse transcription reaction mixture (claims 148-149 and 240) and a kit (claims 150-151 and 241). The process involve the simultaneous amplification of a target viral RNA sequence and reference RNA sequence (claims 114-121, 130-137, 235 and 237) or involve first a simultaneous reverse transcription and then a simultaneous amplification of a target viral RNA sequence and reference RNA sequence (claims 122-129, 138-145, 236 and 238). The reference RNA sequence

of claims 114-151 is a reference RNA sequence that consists of the target viral RNA sequence with a multibase insert into a site within the target viral RNA sequence

Claims 190-234 and 242-248 are directed to processes for quantitation of a target viral RNA sequence in a sample, an amplification reaction mixture (claims 226-228 and 246), a reverse transcription reaction mixture (claims 229-231 and 247) and a kit (claims 232-234 and 248). The process involve the simultaneous amplification of a target viral RNA sequence and reference RNA sequence (claims 190-198, 208-216, 242 and 244) or involve first a simultaneous reverse transcription and then a simultaneous amplification of a target viral RNA sequence and reference RNA sequence (claims 199-207, 217-225, 243 and 245). The reference RNA sequence of claims 190-234 is a reference RNA sequence that comprises a sequence present in the target viral RNA sequence and a sequence that is not present in the target viral RNA sequence.

Support for the Claim Language

As noted above, support for the language that the reference RNA sequence and the (selected) target viral RNA sequence are similar in length can be found in the disclosure of the maxigene in the present application, in which the reference RNA sequence differs from the target viral RNA sequence by the presence of a multibase pair insert into a unique site (page 6, lines 19-21) and by the use of a small insertion in the target viral RNA sequence (page 7, lines 12-15). An example of a multibase pair insert is one of about 20 bases, e.g., a 21 multibase insert (page 6, lines 21-25). Although the specification does not contain an *in haec verba* description of target and reference sequences that are of similar length, the specification does contain an equivalent description of the claimed invention by providing an example of a 21 base multibase insert and the language "small insertion." Such a description is sufficient. *See, Regents of the University of California v. Eli Lilly and Co.*, 119 F.3d 1559, 1566, 43 USPQ2d 1398, 1404 (Fed. Cir.1997) ("[a]n applicant complies with the written description requirement 'by describing the invention, with all its claimed limitations, not that which makes it obvious,' and by using '**such descriptive means as words**, structures, figures, diagrams, formulas, etc., that set forth the claimed invention.'" (emphasis added); *Lockwood v. American Airlines Inc.*, 107 F.3d 1565, 41

USPQ2d 1961 (Fed. Cir. 1997); *Eiselstein v. Frank*, 52 F.3d 1035, 34 USPQ2d 1467 (Fed. Cir. 1995); *Ex parte Holt*, 19 USPQ2d 1211 (Bd Pat App & Inter, 1991) ("It is well established that the invention claimed need not be described *ipsis verbis* in the specification in order to satisfy the disclosure requirements of 35 U.S.C. §112."). *See also*, MPEP §2163.02 where it states that the "subject matter of the claim need not be described literally (i.e. using the same terms or *in haec verba*) in order for the disclosure to satisfy the description requirement". Furthermore, as detailed below, the Examiner has concluded that this language is supported by the present application. In addition, support for the language that the reference RNA and the target viral RNA can be amplified and detected with the same oligonucleotides is supported at page 6, lines 15-18 of the present application.

Summary of the Invention

The present invention is directed to a method for the quantitation of target viral RNA in a sample by simultaneously amplifying a target viral RNA sequence and a known quantity of a reference RNA sequence as an internal standard. That is, the target viral RNA sequence, if present, and the reference sequence are simultaneously amplified in the same reaction mixture. The quantity of target viral RNA present in the sample is determined by comparing the amount of the amplified target viral RNA and the amount of the amplified reference RNA based on the known quantity of reference RNA added as an internal control. The reference RNA sequence may be (a) a reference RNA sequence that consists of the target viral RNA sequence with a multibase insert into a site within the target viral RNA sequence or (b) a reference RNA sequence that comprises a sequence present in the target viral RNA sequence and a sequence that is not present in the target viral RNA sequence. In each instance, the reference RNA sequence and the target viral RNA sequence are of similar length and can be amplified and detected by the same oligonucleotides.

Priority

According to the first paragraph of the specification, the present application is a continuation-in-part of three applications, Serial No. 07/355,296, filed 22 May 1989, Serial No. 07/143,045, filed 12 January 1988 and Serial No. 07/148,959, filed 27 January 1988. Thus, the present application claims priority to each of these three applications. As discussed below, Applicants submit that they are entitled to a priority date of at least 27 January 1988 for the amended claims.

With respect to claims 114-151 and 235-241, Applicants note that these claims are limited to a

> "reference RNA sequence which consists of the selected target viral RNA sequence with a multibase insert into a site within the selected target viral RNA sequence, wherein the reference RNA sequence and the selected target viral RNA sequence are of similar length and can be amplified and detected by the same oligonucleotides."

Thus, the reference RNA sequence with the multibase insert into a site within the selected target viral RNA sequence contains two limitations. The first limitation is that the reference RNA sequence and the selected target viral RNA sequence are of similar lengths. The second limitation is that the reference RNA sequence and the selected target viral RNA sequence can be amplified and detected by the same oligonucleotides. Applicants submit that these limitations are fully supported by the present application and are fully supported by the parent applications. As such, these limitations have an effective priority date of at least 27 January 1988.

With respect to the first limitation, Applicants note that the Examiner has stated that the originally filed application "supports only quantifying target RNA wherein a target and a reference RNA have **similar lengths**." See page 5 of the Office Action mailed 16 March 2006 (emphasis added). This support noted by the Examiner is based on the disclosure in the specification of a maxigene formed by a multibase pair insert into a unique site in the target sequence (pages 4-5 of the Office Action mailed 16 March 2006) and a small insertion cloned into a viral RNA as an internal standard (page 5 of the Office Action mailed 16 March 2006). On the basis of these disclosures, the Examiner concluded that there is no support for a "reference RNA significantly different in length than the target." See, page 5 of the Office

Action mailed 16 March 2006. In addition, the Examiner has stated that "the original disclosure does not support using a primer pair for amplifying both RNAs [a reference RNA and a target RNA] wherein the amplification substrates (RNAs) are significantly different." See, page 6 of the Office Action mailed 16 March 2006. If the specification doesn't support RNAs of different lengths, it must support RNAs of similar lengths. In fact, the Examiner stated that "the RNAs amplified by the disclosed primers are similar in length" in order for the reference RNA to be amplified and detected by the same oligonucleotides as the target. See, page 6 of the Office Action mailed 16 March 2006. Thus, and as the Examiner has concluded, the present application fully supports the limitation with respect to a reference RNA sequence that is similar in length to the target viral RNA sequence.

The disclosure of a "'maxigene' formed by a mutli-base pair insert into a unique site" and "a small insertion" in a target sequence are fully supported in the parent applications. For example, the '045 application discloses a reference RNA sequence with a small insertion. Specifically, the '045 application discloses a small insertion in a segment of the HCMV major IE gene IE1 and its use as an internal control for quantifying the amount of HCMV present in a sample. See, page 8, lines 10-18 of the '045 application. The small insertion is specifically disclosed at page 8, lines 12-15 of the '045 application. In addition, the '959 application discloses a "'maxigene' formed by a multi-base insert into a unique site" for quantitation of viral levels in patient samples. See, page 3, lines 23-28 of the '959 application. According to the '959 application, one example of such an insert for use in quantifying HIV-1 is a multibase insert into the unique KpnI site of the 3' ORF region. See, page 3, lines 28-29 of the '959 application. The '959 application discloses that a preferred reference RNA includes a 22 base pair insert (page 3, lines 29-30 of the '959 application) while an example of a multibase insert has 21 nucleotides (page 3, line 32 of the '959 application). In addition, the '959 application discloses that the reference RNA can be amplified and detected by the same oligonucleotides as the target. See, page 3, lines 26-26. As detailed in the preceding paragraph above, the Examiner has stated that the present application has support for a reference RNA sequence with a multibase insert in the target sequence in which the reference RNA sequence and the target sequence are similar in

length based on the disclosures in the present application. As detailed in this paragraph, the '045 application and the '959 application contain the same disclosures as found in the present application. Thus, Applicants submit that the parent applications fully support a reference RNA sequence with a multibase insert in the target sequence in which the reference RNA sequence and the target viral RNA sequence have similar lengths. Consequently, this limitation in the claims has priority to U.S. patent application Serial Nos. 07/143,045 and 07/148,959, with priority dates of 12 January 1988 and 27 January 1988, respectively.

With respect to the second limitation, Applicants note that the Examiner has stated that "the specification discloses that '[a]n additional aid to quantitation of virus levels in patient samples is provided by a reference RNA which can be amplified and detected by the same oligonucleotides used for authentic virus RNA samples.'" See, page 6 of the Office Action mailed 16 March 2006 (emphasis in original). Thus, the Examiner has concluded that there is support in the present application for this second limitation. The disclosure that the reference RNA sequence and the target viral RNA sequence can be amplified by the same oligonucleotides is fully support in a parent application. Specifically, page 3, lines 23-26 of the '959 application states "[a]n additional aid to quantitation of virus levels in patient samples is provided by a reference RNA which **can be amplified and detected by the same oligonucleotides** used for authentic virus RNA samples." (emphasis added) This disclosure in the '959 application is identical to the disclosure in the present application. Thus, Applicants submit that the parent application fully supports a reference RNA sequence and a target viral RNA sequence which can be amplified by the same. Consequently, this limitation in the claims has priority to U.S. patent application Serial No. 07/148,959, with priority date 27 January 1988.

Thus, Applicants submit that the priority dates for the subject matter claims 114-151 and 235-241 are the filing dates of the '045 and '959 applications, i.e., 12 January 1988 and 27 January 1988.

With respect to claims 190-234 and 242-248, Applicants note that these claims are limited to a

> "reference RNA sequence comprises a sequence present in the selected target viral RNA sequence and a sequence not present in the selected target viral

> RNA sequence, wherein the reference RNA sequence and the selected target viral RNA sequence are of similar length and can be amplified and detected by the same oligonucleotides."

As detailed above, Applicants note that maxigene contains an insert in the target viral RNA sequences (e.g., see page 3, lines 27-28 of the '959 application). Because the insert sequence is not a target viral RNA sequence, the "maxigene" comprises target viral RNA sequence and non-target viral RNA sequence. The amplified target viral RNA sequence and the amplified reference RNA sequence are distinguishable by size. See, page 4, lines 13-14 of the '959 application. Alternatively, the amplified target viral RNA sequence and the amplified reference RNA sequence are distinguishable by probes. See, page 4, lines 15-19 of the '959 application. Reference RNA sequences with a multibase insert, i.e., reference RNA sequences that contain target and non-target viral RNA sequences, are capable of being distinguished from the target viral RNA sequence by either size or by probes. Thus, the '959 application fully supports a reference RNA sequence that contains target viral RNA sequence and non-target viral RNA sequence. A similar reference sequence is shown in the 'the '045 application, i.e., a small insertion (page 8, lines 12-15 of the '045 application). As detailed above, the reference RNA sequence, which can be expressed as either (i) a reference RNA sequence with a multibase insert in the target viral RNA sequence or (ii) a reference sequence which comprises a sequence present in the selected target viral RNA sequence and a sequence not present in the selected target viral RNA sequence, and the target viral RNA sequence have similar lengths. Consequently, the first limitation in these claims has priority to U.S. patent application Serial Nos. 07/143,045 and 07/148,959, with priority dates of 12 January 1988 and 27 January 1988, respectively. The Examiner has also indicated that there is support in the present application for the limitation that the reference RNA sequence and the target viral RNA sequence can be amplified and detected by the same oligonucleotides. This same disclosure is found in one of the parent applications, e.g., see page 3, lines 23-26 of the '959 application. Consequently, this second limitation in these claims has priority to U.S. patent application Serial No. 07/148,959, with priority date 27 January 1988. Therefore, Applicants submit that the subject matter of claims 190-234 and 242-248 has priority and benefit to the '045 and '959 applications for the

same reasons that the subject matter of claims 114-151 and 235-241 has priority and benefit to the '045 and '959 applications.

Thus, Applicants submit that the priority dates for the subject matter claims 190-234 and 242-248 are the filing dates of the '045 and '959 applications, i.e., 12 January 1988 and 27 January 1988.

New Matter Rejection

In the Office Action mailed 16 March 2006, the Examiner rejected claims 114-234 under 35 U.S.C. § 112, first paragraph for new matter, i.e., a reference RNA sequence of any size. Applicants submit that the amendment of the claims to specify that the reference RNA sequence and the target viral RNA sequence are similar in length and can be amplified and detected by the same oligonucleotides obviates this rejection obviates this rejection. Applicants submit that the amended claims no longer encompass any size reference RNA sequence. Applicants further submit that the application clearly discloses quantitation of a target viral RNA sequence using a reference sequence which has a similar length as the target viral RNA sequence and which can be amplified and detected by the same oligonucleotides as stated by the Examiner in the Office Action mailed 16 March 2006 and as detailed above.

In view of the amendments to the claims and the above remarks, it is submitted that claimed subject matter is fully described in the specification. Withdrawal of this rejection is requested.

Enablement Rejection

In the Office Action mailed 16 March 2006, the Examiner rejected claims 114-234 under 35 U.S.C. § 112, first paragraph for lack of enablement for the scope of the claimed subject matter, i.e., for a reference RNA sequence of any size. Applicants submit that the amendment of the claims to specify that the reference RNA sequence and the target viral RNA sequence are similar in length and can be amplified and detected by the same oligonucleotides obviates this rejection. Applicants submit that the amended claims no longer encompass any size reference

RNA sequence. Applicants further submit that the application clearly discloses quantitation of a target viral RNA sequence using a reference sequence which has a similar length as the target viral RNA sequence and which can be amplified and detected by the same oligonucleotides as stated by the Examiner in the Office Action mailed 16 March 2006 and as detailed above.

In view of the amendments to the claims and the above remarks, it is submitted that the claimed subject matter is fully enabled by the specification. Withdrawal of this rejection is requested.

Anticipation Rejection

In the Office Action mailed 16 March 2006, the Examiner rejected claims 146-151 and 229-234 under 35 U.S.C. § 102(a) as being anticipated by Murakawa et al. (*DNA* 7:287-295, 1988). As detailed above, Applicant submits that the amended claims are entitled to the priority dates of the '045 and '959 applications, i.e., 12 January 1988 and 27 January 1988. These dates are prior to the date of Murakawa et al., and thus Murakawa et al. is not prior art against the amended claims.

In view of the amendments to the claims and the above remarks, it is submitted that the claimed subject matter is entitled to priority dates of 12 January 1988 and 27 January 1988 and is not anticipated by Murakawa et al. Withdrawal of this rejection is requested.

Obviousness Rejections

In the Office Action mailed 16 March 2006, the Examiner rejected claims 115-145 and 190-228 under 35 U.S.C. § 103(a) for being obvious over Murakawa et al. in view of Chelly et al. (*Nature* **333**:858-860, 1988). As detailed above, Applicant submits that the amended claims are entitled to the priority dates of the '045 and '959 applications, i.e., 12 January 1988 and 27 January 1988. These dates are prior to the dates of Murakawa et al. and Chelly et al. Thus, neither Murakawa et al. nor Chelly et al. is prior art against the amended claims.

In view of the priority date to which the amended claims is entitled, it is submitted that the claimed subject matter is not obvious over the cited references. Withdrawal of this rejection is requested.

Furthermore, Applicants submit that the amended claims are not obvious from the cited references. Specifically, the Examiner contends that Murakawa et al. discloses (a) selecting a sequence present in a target viral RNA (Fig. 1), (b) a reference RNA consisting of the selected target and an insertion of 21 bases (page 292; Figs. 7 and 8), (c) adding a known quantity of a reference RNA to a sample (50 ng: page 293, Fig. 8; equimolar amount: page 292) and (d) simultaneous amplification of target and reference RNA. The Examiner notes that Murakawa et al. does not specifically disclose measuring the amount of RNA and determining the amount of target RNA before amplification from the amount of reference RNA. The Examiner contends that these limitations are disclosed by Chelly et al. Applicants submit that the Examiner is in error in this rejection.

Murakawa et al. discloses a process in which an equal amount of target viral RNA and a reference RNA which is a target viral RNA with an insert are added to an amplification reaction mixture (page 292; Fig.7). Hence a known amount of both the target viral RNA and reference RNA are present in the amplification reaction mixture. The target viral RNA and reference RNA are then simultaneously amplified for 12 rounds (page 292; Fig. 7). A sample of the amplification reaction mixture is then removed and a transcription reaction is performed on the sample (page 292; Fig. 7). The transcription reaction products are then electrophoresed and the intensities of the bands compared (Fig. 7). Neither the amounts of the amplified products are measured nor the amount of original target viral RNA is determined. In this process, the amount of the target viral RNA that was originally present was known and did not need to be determined.

Murakawa et al. also discloses a similar process in which a sample suspected of containing a target viral RNA is amplified (Fig. 8). After 15 rounds of amplification, a known amount of the reference RNA is added to a portion of the sample containing the amplified target viral RNA (Fig. 8). This new sample is subjected to 10 rounds of amplification in which the target RNA and reference RNA are simultaneously amplified (Fig. 8). A sample of the

amplification reaction mixture is then removed and a transcription reaction is performed on the sample (page 292; Fig. 8). The transcription reaction products are then electrophoresed and the intensities of the bands compared (Fig. 8). Neither the amounts of the amplified products are measured nor the amounts of original target viral RNA are determined. In this process, two separate amplification reactions are conducted, one without the reference RNA and one with the reference RNA. In both of the process disclosed by Murakawa et al. in which a reference RNA is added, an amplification reaction is performed and a transcription reaction is performed on the amplified reaction product. Murakawa et al. does not describe the simultaneous amplification of a target RNA and a reference RNA without a transcription reaction before electrophoresis of the amplified products.

In the Office Action mailed 16 March 2006, the Examiner contended that Chelly et al. discloses adding a reference RNA to a sample and that quantitation of the DNA of interest is determined. The Examiner is incorrect in her reading of this reference. Chelly et al. describes the simultaneous amplification of an endogenous, heterologous reference sequence, i.e., aldolase A, and a target sequence, i.e., dystrophin. Instead of adding a reference RNA to a sample, the sample that is amplified by Chelly et al. contains both dystrophin and aldolase A. Chelly et al. clearly does not add the aldolase A (reference sequence) to the sample. Because Chelly et al. does not add a reference RNA, it does not add a known quantity of the reference RNA. Thus, the Examiner's interpretation of an added reference RNA in Chelly et al. is incorrect. Applicants also note that this interpretation is counter to facts found by the Board in the Wang v. Murakawa interference (Patent Interference No. 105,055; see Memorandum Opinion and Order (Decision on Wang preliminary motion 1) dated 5 November 2003).

In addition, Applicants submit that Chelly et al. does not describe a process for quantitation of a target sequence. Chelly et al. specifically states that the absolute value of dystrophin mRNA cannot not be determined directly but that a relative figure can be obtained by comparison with the aldolase A internal standard. See, Figure 3 of Chelly et al. The internal standard of Chelly et al. is a standard that is present in the samples used for amplification and is not a standard that is added to the sample in a predetermined amount for simultaneous

amplification and quantitation. The amount of the internal standard that is present is not known, and thus, a ratio of the amplified products is determined from which a ratio of starting mRNAs is calculated. Chelly et al. does not described the determination of an absolute amount of the target sequence that is present in the sample which is in direct contrast to the present invention's quantitation of the target sequence present in the sample.

More specifically, Chelly et al. also does not describe a method for the precise quantitation of a target RNA sequence, but merely describes a method for the crude estimation of the relative amount of the target dystrophin mRNA. Chelly et al. explains that aldolase A mRNA was used to check efficiency of the amplification reaction. See page 858, left column ("To check the efficiency of the procedure, we simultaneously co-reverse transcribed and co-amplified another transcript as an internal standard in the same test tube."). The calculations using the "labeled primers" to determine the relative amount of the target dystrophin mRNA in the sample (relative to total mRNA) were based on the ratio of target to standard and the estimated amount of standard mRNA in certain specific tissues. See page 859, left column ("From this value we deduced the amount of dystrophin mRNA relative to total mRNA (Table 1)."). In this deduction, Chelly et al. provides, at best, an estimate that the amount of aldolase A mRNA ranges from about "0.1-0.5% of total mRNA in skeletal muscle, and at least ten times less in other tissues." See page 859, left column. The title of Table 1 ("Estimation ...), as well as the abstract ("quantitative estimate"), confirms that Chelly et al. only estimated the amount of dystrophin mRNA in the sample because it only estimated the amount of the aldolase A internal standard that was present in the tissue samples. Although Chelly et al. describes a ratio for determining an amount of target sequence that may be present as stated by the Examiner in the Advisory Action mailed 4 October 2006, the ratio cannot provide a quantitation of the amount of target sequence present **because the amount of the reference sequence in the tissue samples is merely estimated**. Applicants submit that a quantitative estimate is not quantitation and no quantitative amount is provided by Chelly et al. Chelly et al. also states that the absolute value of dystrophin mRNA can not be determined directly but a relative figure can be deduced. See legend to Figure 3 under "Methods." Chelly et al. requires the use of ratios to measure

expression of the target dystrophin mRNA because the standard, aldolase A, is not used in a known initial amount, as required by the present invention. It is the use of a known quantity of the reference RNA sequence in the claimed method, an element missing from Chelly et al., which enables the quantitation of target viral RNA, because the initial amount of the reference sequence is known.

The labeled primers were used to determine the efficiency of the reaction and to calculate the ratio of dystrophin mRNA to aldolase A mRNA in the sample. See, page 859, top of left column. The labeled primers were not used to quantify the amount of target RNA in the sample. This calculation only provides a ratio of dystrophin mRNA to aldolase RNA and does not provide any quantitation of the dystrophin mRNA. The ratio is useful in identifying tissues that express dystrophin.

From the above analysis of Chelly et al., it is apparent that Chelly et al.'s experimental objective was to provide a relative estimate of the amount mRNA present in a sample, such that the estimate was, at best, a comparative estimate. There is no attempt to use PCR to quantitate actual amounts of target. This objective differs from Applicants' objective which was to use PCR to quantitate a target viral RNA. Because Chelly et al.'s objective differs from Applicants' objective, it is evident that Chelly et al. could not motivate or suggest the claimed invention even if the critical elements of the present invention were disclosed therein.

Chelly et al. only discloses the determination of a relative quantity of a target RNA sequence. It does not disclose the absolute quantitation of the target sequence. Because Chelly et al. does not describe such quantitation, it does not supply the element missing from Murakawa et al. that has been noted by the Examiner. Thus, Applicants submit that the combination of Murakawa et al. and Chelly et al. does not render the invention obvious.

Finally, Applicants submit that there is no motivation or suggestion to combine the prior art in the manner proposed by the Examiner to arrive at the presently claimed subject matter. According to the Examiner, Murakawa et al. discloses a reference sample that is a target sequence with an insert that is similar in length as the target sequence. Chelly et al. discloses a reference sequence which is totally heterologous to the target sequence, i.e., the reference

sequence that Chelly et al. uses contains no sequence found in the target sequence. Thus, Applicants submit that there would be no motivation to combine Chelly et al. with Murakawa et al.

Furthermore, even if there was motivation to combine the secondary reference Chelly et al. with the primary reference Murakawa et al., the combination would not yield the claimed subject matter. Specifically, as described above, Murakawa et al. teaches two processes in which a reference RNA sequence is used. In the first process, a reference RNA sequence and a target sequence are added to a reaction mixture and simultaneously amplified for 12 rounds of amplification. A transcription reaction is then performed on the amplified products before the amplified products are analyzed. In the second process, a sample is amplified for 15 rounds and then a reference RNA sequence is added to the mixture. This mixture is then amplified for 10 rounds during which the amplified target sequence and the reference RNA sequence are simultaneously amplified. A transcription reaction is then performed on the amplified products before the amplified products are analyzed. The Examiner relies on Chelly et al. for the steps of measuring the amount of amplified products and quantifying a target RNA. The addition of these steps of Chelly et al. to the processes of Murakawa et al. would yield either (i) a process in which a known amount of target viral RNA sequence and a known amount of a reference RNA sequence are amplified, the amplified products are transcribed and the transcribed products are measured and quantified or (ii) a process in which the target viral RNA sequence is amplified prior to the addition of the reference RNA sequence, the amplified target viral RNA sequence and reference RNA sequence are amplified, the amplified products are transcribed and the transcribed products are measured and quantified. Neither of these processes describes or suggests the claimed subject matter. Consequently, a combination of the prior art as proposed by the Examiner does not render the claimed subject matter obvious.

In view of the amendments to the claims and the above remarks, it is submitted that the claims are not obvious over the combination of Murakawa et al. in view of Chelly et al. Withdrawal of this rejection is requested.

The Examiner also rejected claims 152-189 under 35 U.S.C. § 103(a) for being obvious over Murakawa et al. in view of Chelly et al. further in view of Arya (*Proc Natl Acad Sci USA* **84**:5429-5433, 1987). Applicants submit that the cancellation of these claims obviates this rejection. Withdrawal of this rejection is requested.

Advisory Action

In the Advisory Action mailed 4 October 2006, the Examiner asserted that the "newly added limitation 'wherein the reference RNA sequence and the selected target viral RNA sequence are of similar lengths and capable of being amplified by the same oligonucleotides' raises new issues under 35 USC 102, 103, 112, first and second paragraphs." This limitation had been added in the Amendment After Final filed on 18 September 2006, and this amendment was not entered. A similar limitation is set forth in the presently amended claims. The limitation now reads "wherein the reference RNA sequence and the selected target viral RNA sequence are of similar length and can be amplified and detected by the same oligonucleotides."

With respect to any potential new issue under 35 U.S.C. §§ 102 and 103 alluded to by the Examiner, Applicants are not aware of any prior art in addition to that previously considered by the Examiner during the prosecution of this application. As detailed above, the claimed subject matter is entitled to the priority dates of the '045 and '959 applications, i.e., 12 January 1988 and 27 January 1988. These dates antedate the dates of many of the references in the art of quantitative PCR as shown herein. In addition, either the date of invention antedates other references or the subject matter described in other references is the invention of the present Applicants. Both of these facts have been established by previously filed declarations.

Finally, the claimed subject matter is not the same subject matter as the claims of the Wang et al. patents (US 5,219,727 and US 5,476,774). Applicants submit that this issue was addressed and determined by the Board of Patent Appeals and Interferences ("the Board") in Patent Interference No. 105,055. Specifically, in its Memorandum Opinion and Order (Decision on Wang preliminary motion 1) dated 5 November 2003, the Board concluded that a reference RNA which can be amplified and detected by the same oligonucleotides used for authentic virus

RNA samples, including a reference RNA containing a multibase insert, does not necessarily require or result in the use of a shared primer pair as required by the Wang et al. patents. Because such a reference RNA did not necessarily require or result in the use of a shared primer pair, the Board concluded that the claims of Murakawa et al. that did not specify a shared primer pair but only specified a reference RNA and target viral RNA that can be amplified and detected by the same oligonucleotides did not claim the same invention as the Wang et al. patents. In view of this decision by the Board, Applicants submit that the claimed subject matter of the present application is not the same as the claimed subject matter of the Wang et al. patents. As discussed above, the priority dates for the presently claimed subject matter are the filing dates of the '045 and '959 applications, i.e., 12 January 1988 and 27 January 1988. These dates are prior to the earliest filing date of both Wang et al. patents, and thus these patents are not prior art. In addition, Applicants have sworn behind theses patents in a previously filed declaration.

With respect to any potential new issues under 35 U.S.C. § 112, first paragraph alluded to by the Examiner, Applicants submit that the newly added limitations are fully supported by the present application as detailed above and as stated by the Examiner in the Office Action mailed 16 March 2006. Thus, Applicants submit that there is no issue of new matter under 35 U.S.C. § 112, first paragraph for the newly added claim limitations.

In addition, Applicants have fully described a reference RNA sequence that either (i) consists of the selected target viral RNA sequence with a multibase insert into a site within the selected target viral RNA sequence or (ii) comprises a sequence present in the selected target viral RNA sequence and a sequence not present in the selected target viral RNA sequence. For either reference RNA sequence, the reference RNA sequence and the target viral RNA sequence are of similar length and can be amplified and detected by the same oligonucleotides. Applicants have specifically described in the specification the quantitation of target viral RNA using a reference RNA in which the two can be amplified and detected by the same oligonucleotides. See, page 6, lines 15-18. Applicants have also described reference RNA sequences that contain an insert in the target viral RNA sequence, either as a multibase insert or as a small insert. See, page 6, lines 19-25 and page 7, lines 12-15. In either instance, the reference RNA sequence is of

similar length to the target viral RNA sequence. In addition, in either instance the reference RNA sequence comprises target viral RNA sequence and non-target viral RNA sequence. The specification clearly discloses that virus levels in patients can be quantified for any virus. See, page 6, lines 15-18. Two specific examples are provided, namely HIV-1 and HCMV. See pages 6 and 7. It is evident from this analysis of the specification that newly added limitations have been disclosed in descriptive words. These descriptive words clearly show that Applicants were in possession of the invention with all of the claimed limitations. *Regents of the University of California*, 119 F.3d at 1566, 43 USPQ2d at 1404. Thus, Applicants submit that there is no issue of written description under 35 U.S.C. § 112, first paragraph for the newly added claim limitations.

Furthermore, Applicants submit that the specification fully enables the presently claimed subject matter in light of the newly added limitations. Specifically, Applicants note that the Examiner has stated that the specification enables the method in which the amplified portions of the target viral RNA and a reference RNA are similar in length and can be amplified and detected by the same oligonucleotides. See, pages 7-9 of the Office Action mailed 16 March 2006. The claim limitations clearly exclude insertions of unspecified length which the Examiner has contended is not enabled. See, page 8 of the Office Action mailed 16 March 2006. The art cited by the Examiner on page 8, particularly Murakawa et al. and Zaia et al., which were published subsequent to the earliest effective priority dates for the presently claimed subject matter (as detailed above), clearly show that the claimed process is enabled for reference RNA sequences that are similar in length to the target viral RNA sequences and that no undue experimentation was required to practice the claimed invention.

The specification provides guidance for accomplishing quantitation of virus levels through the use of reference RNA sequence that can be amplified and detected by the same oligonucleotides as the target viral RNA sequence and through the use of reference RNA sequences that are similar in length (i.e., a small insert or a multibase insert as illustrated by an insert of 21 bases) to the target viral RNA sequences.

Furthermore, the Examiner has not provided any evidence that primers that vary only slightly, such as by a single nucleotide, could not be used to amplify the reference RNA sequence as efficiently as the original primers amplify the target viral RNA sequence. Although primer pairs that differ in this manner are not the same primer pair, a skilled artisan would reasonably predict that the efficiency of amplification would be similar for the reference RNA sequence as for the target viral RNA sequence in this instance, particularly in the absence of any evidence to the contrary. There would be no undue experimentation to determine if the amplification reactions proceed as efficiently using such slightly different primers and could be performed by routine screening. Thus, Applicants submit that a skilled artisan can readily practice the claimed invention for these types of primers.

In addition, Applicants note that the Examiner has indicated that the level of skill in the art is high. See, page 8 of the Office Action mailed 16 March 2006. In view of this high level of skill in the art and the guidance provided in the specification, it is evident that a skilled artisan can readily make and use the claimed invention without undue experimentation, especially as illustrated by the post-filing literature cited by the Examiner. Thus, Applicants submit that there is no issue of enablement under 35 U.S.C. § 112, first paragraph for the newly added claim limitations.

With respect to any potential new issues under 35 U.S.C. § 112, second paragraph alluded to by the Examiner, Applicants submit that the newly added limitations are definite to a skilled artisan. Definiteness is determined with reference to a person of ordinary skill in the art. *Miles Laboratories, Inc. v. Shandon Inc.*, 997 F.2d 870, 875, 27 USPQ2d 1123, 1126 (Fed. Cir. 1993), *cert. denied*, 510 U.S. 1100 (1994) ("The test for definiteness is whether one skilled in the art would understand the bounds of the claim when read in light of the specification."); *In re Warmerdam*, 33 F.3d 1354, 1361, 31 USPQ2d 1754, 1759 (Fed. Cir. 1994) ("The legal standard for definiteness is whether a claim reasonably apprises those of skill in the art of its scope.").

Applicants submit that a skilled artisan knows what the term "of similar length" means, especially when read in the context of the claims and the specification. The claims specifically recite that "the reference RNA sequence and the selected target viral RNA sequence are of

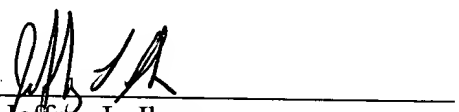
the same oligonucleotides" refer, he understands the metes and bounds of these claim terms. Therefore, Applicants submit that the claims are definite to a skilled artisan. *Miles Laboratories*, 997 F.2d at 875, 27 USPQ2d at 1126; *In re Warmerdam*, 33 F.3d at 1361, 31 USPQ2d at 1759. Thus, Applicants submit that there is no issue of definiteness under 35 U.S.C. § 112, second paragraph for the newly added claim limitations.

Concluding Comments

In view of the above amendments and remarks, it is submitted that the claims are fully supported by the instant application, entitled to a priority date of at least 27 January 1988 and are patentable over the prior art of record. Reconsideration of this application and early notice of allowance is requested. The Examiner is invited to telephone the undersigned if it will assist in expediting the prosecution and allowance of the instant application.

Respectfully submitted,

ROTHWELL, FIGG, ERNST & MANBECK, p.c.

By 

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